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Jacob Cantrell
Washington University in St. Louis

Driving Gene Expression in the Heterochromatic Environment of the Fourth Chromosome of *D. melanogaster*

Jacob Cantrell

Mentor: Sarah Elgin

Genomes of higher eukaryotes can be divided into two fundamental and dynamic subtypes: euchromatin and heterochromatin. The fourth chromosome of *Drosophila melanogaster* is of particular interest because, despite the fact that it appears to be entirely heterochromatic, the genes within this densely packaged chromosome are expressed. Genes that are active in a euchromatic environment are silenced when transposed to heterochromatin. Our aim is to identify heterochromatic gene regulatory elements that drive their active transcription.

Insertion of a euchromatic *hsp70-white* transgene, which exhibits a uniform red eye phenotype when present in euchromatin, into heterochromatic regions on the fourth chromosome results in sporadic silencing, or Position Effect Variegation (PEV). We replaced the *hsp70* promoter of *hsp70-white* with a genomic fragment of a highly expressed fourth chromosome gene, *Rad23*. The fragment includes the *Rad23* promoter region and ~1 kb of an upstream sequence. Insertion of the *Rad23-white* transgene into the same location on the fourth chromosome switched the *hsp70-white* PEV phenotype to a uniform red eye, suggesting that the *Rad23* fragment is sufficient to drive strong expression of the euchromatic *white* reporter.

A series of transgenic reporter constructs containing fragments of varying lengths of the *Rad23* promoter region were prepared by molecular cloning and injected into embryos of the *Drosophila melanogaster* test line. Results show that the promoter region of *Rad23* containing 100 base pairs upstream of the TSS is sufficient to achieve full expression of *white* in a heterochromatic environment. However, a smaller fragment (only 50 bp upstream of the TSS) resulted in a fly with a completely white eye, indicating the loss of a key promoter element. Additional experiments to narrow down what elements of the noncoding regulatory region drive the expression of the reporter gene, and what constructs produce a PEV phenotype, are underway.